

2012 Mixture Interpretation Workshop:

Mixtures Using *SOUND* Statistics, Interpretation, & Conclusions



Different Assumptions- Different Interpretations

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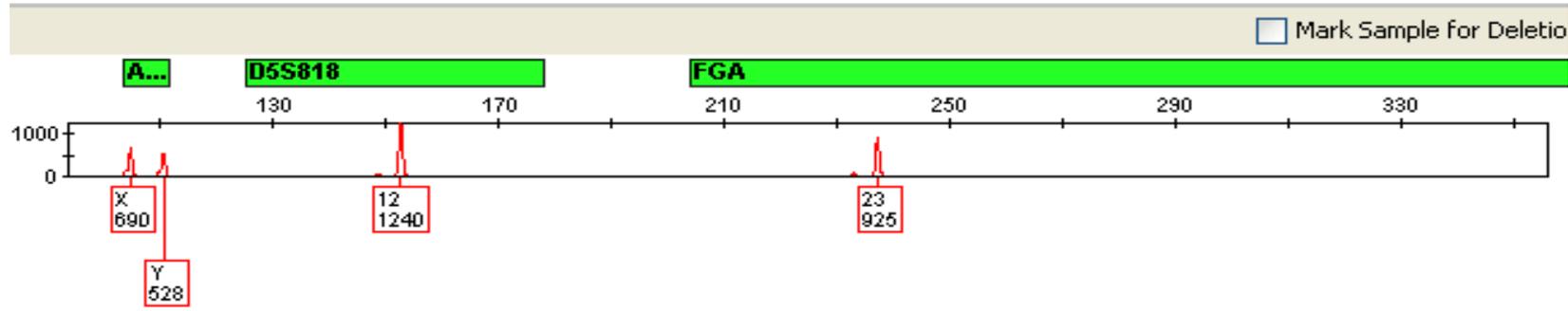
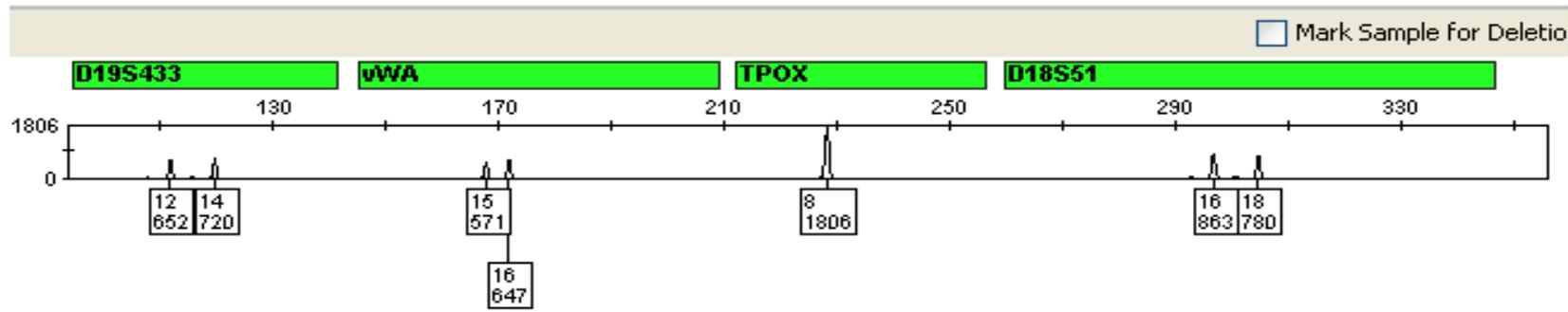
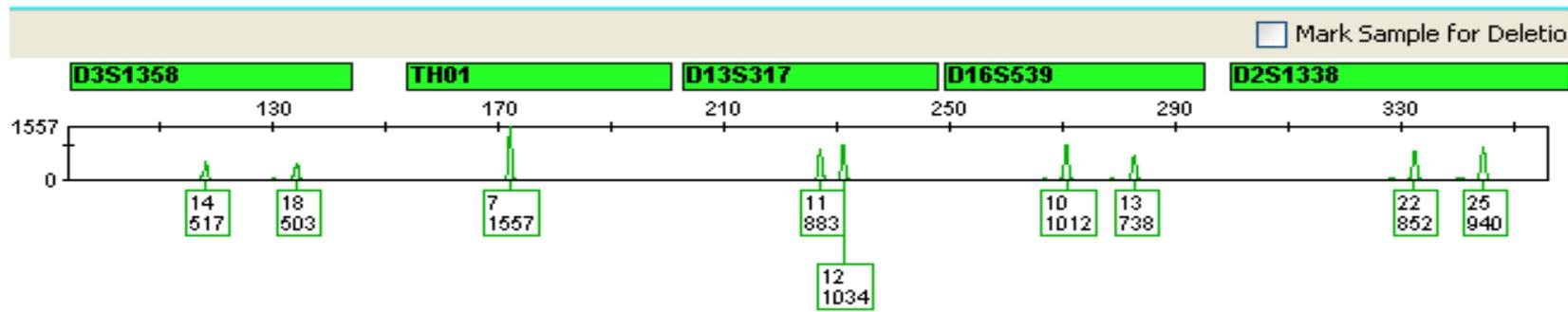
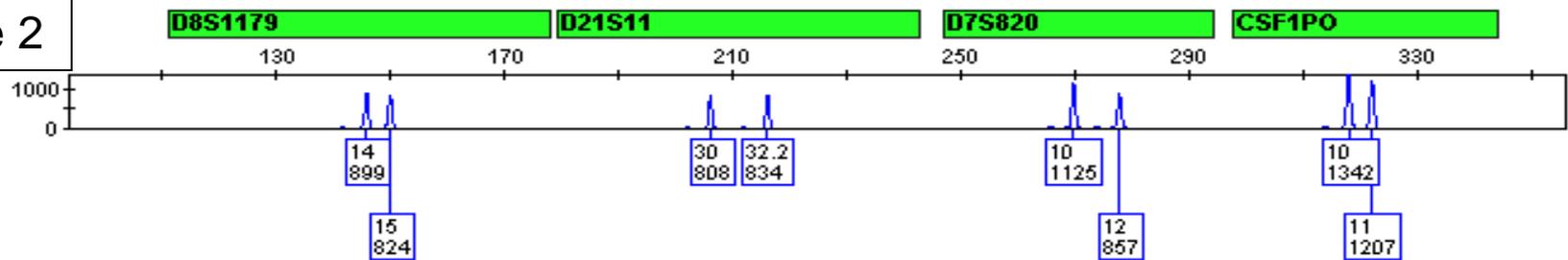
October 15, 2012

Nashville, TN



Profile 2

Single Source Sample



Single Source Sample

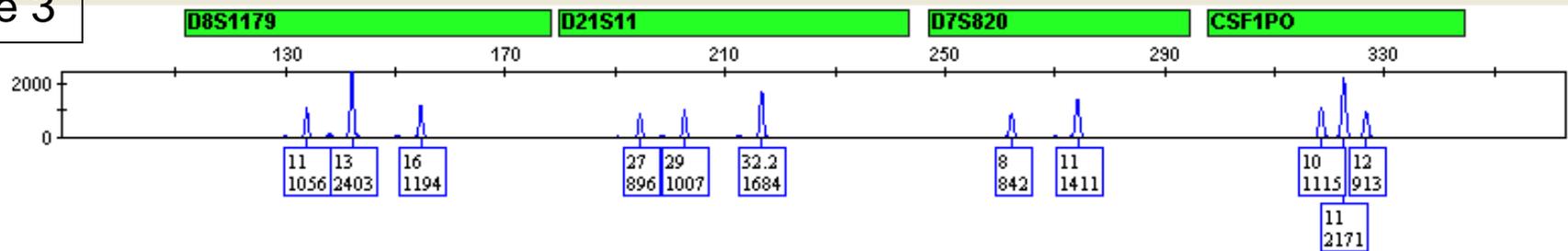


High Certainty Leads to High Confidence

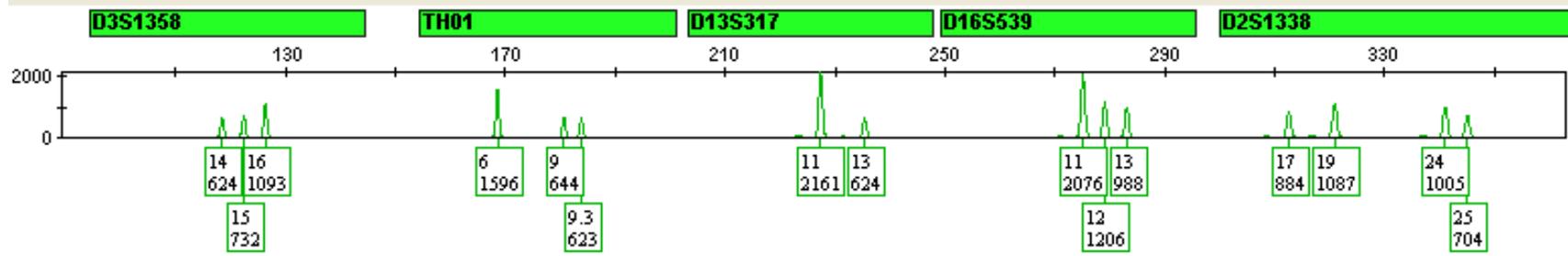
- **High peaks** above stochastic threshold
 - **True alleles** easy to distinguish from stutter & other artifacts
 - **All alleles present** with high confidence
- **1 or 2 alleles** at all loci
- **Balanced peak heights** (inter- & intra-locus)
 - **Number of Contributors** easily assumed
 - Single source
 - **Genotypes** easy to determine

Profile 3

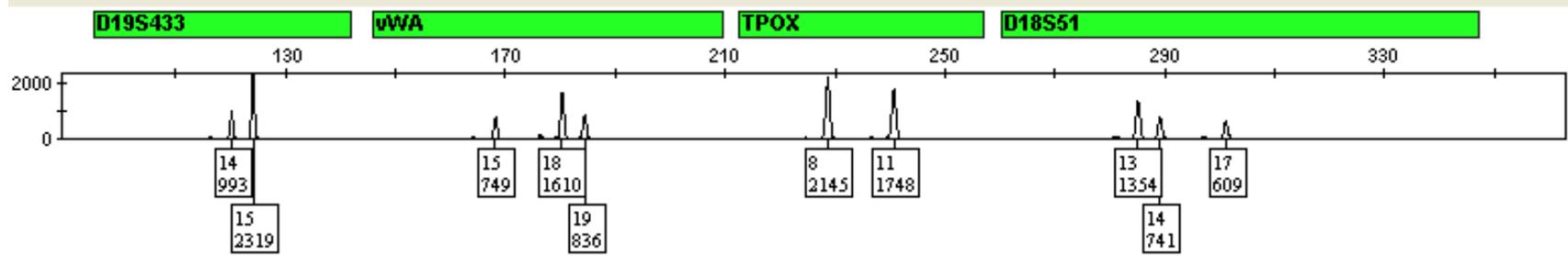
Two-Person Mixture



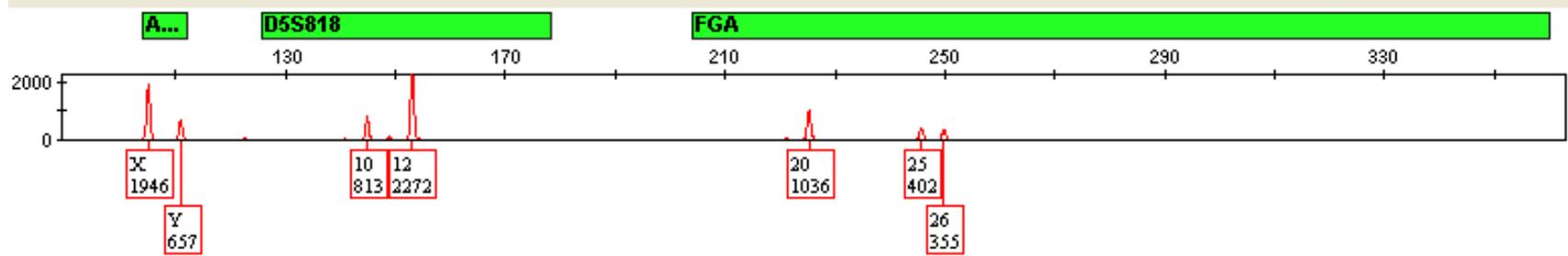
Mark Sample for Deletion



Mark Sample for Deletion



Mark Sample for Deletion



Two-Person Mixtures



High Certainty Leads to High Confidence

- **High peaks** above stochastic threshold
 - **True alleles** easy to distinguish from stutter & other artifacts
 - **All alleles present** with high confidence
- **≤ 4 alleles** at all loci
 - **Number of Contributors** reasonably assumed as 2
 - **Peak heights** consistent with 2 person mixture

Two-Person Mixtures



High Certainty Leads to High Confidence

- **Genotypes and Genotype combinations**
 - Limited number of possible genotypes present
 - Easy to determine
- **May increase certainty of genotypes by:**
 - Assessing mixture ratio
 - Determining if distinguishable/major:minor or indistinguishable mixture
 - Deducing second contributor if one contributor is known

REAL Casework



Increased uncertainty and decreased confidence for some profiles

With that comes increased chance/risk that different interpretations may come from:

- Other analysts in your lab
- Other experts outside of lab

Change in assumptions regarding data affects the interpretation

REAL Casework Experience



Situations with **increased uncertainty, and therefore decreased confidence:**

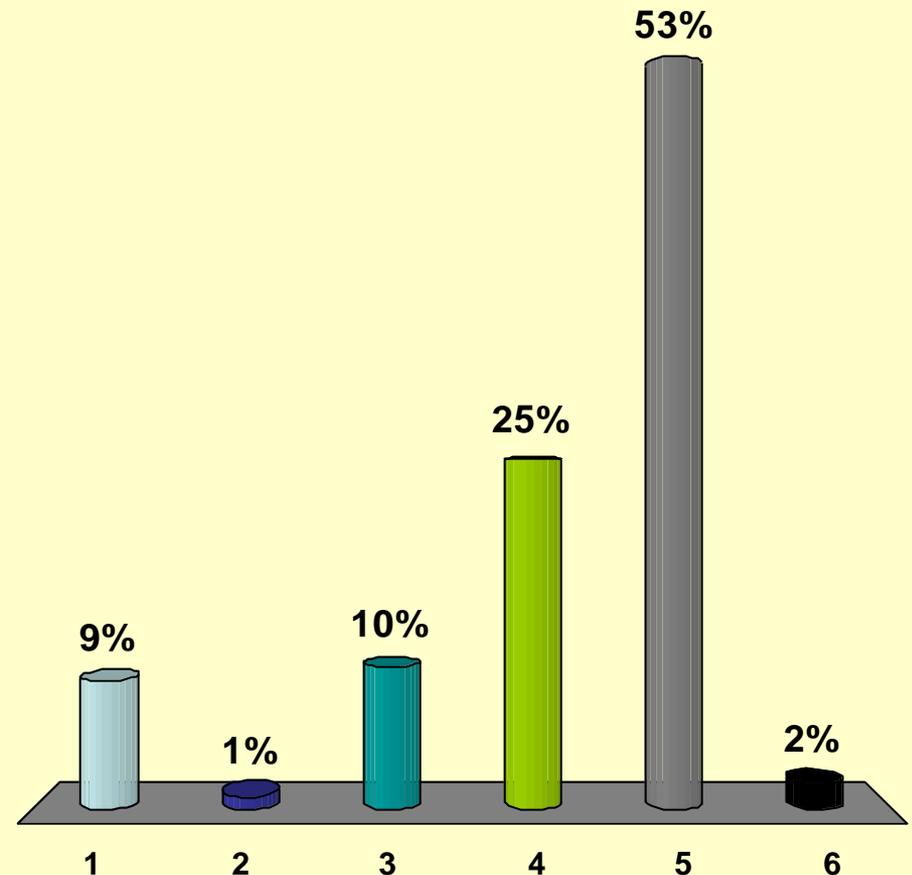
- Alleles vs. artifacts? (LT or high level DNA)
- Stochastic effects possible? (Low peak heights; all or some below stochastic threshold)
 - Sure all alleles are present (drop-out)?
 - Elevated stutter & drop-in present?
- Number of contributors? 1, 2, 3 or more?
- Inability to confidently associate all alleles into reasonable genotypes

When there are different opinions for reporting a profile, our lab reports...

1. Inconclusive
2. Conclusion most favorable to client
3. Conclusion most agree on
4. Single consensus agreement
5. What the technical reviewer/leader says
6. All conclusions with different assumptions

Data from 109 responses

ISHI Mixture Workshop (Oct 2012)



What do you do when...



You have increased uncertainty, and therefore decreased confidence?

Options for interpreting and reporting:

1. Do not interpret the data → report inconclusive
 - When uncertainty is too high
2. Pick one interpretation to report
 - When have minimal uncertainty
3. Interpret and report the data under two or more different assumptions
 - When certainty is medium-to-high but possible scientifically sound alternatives exist

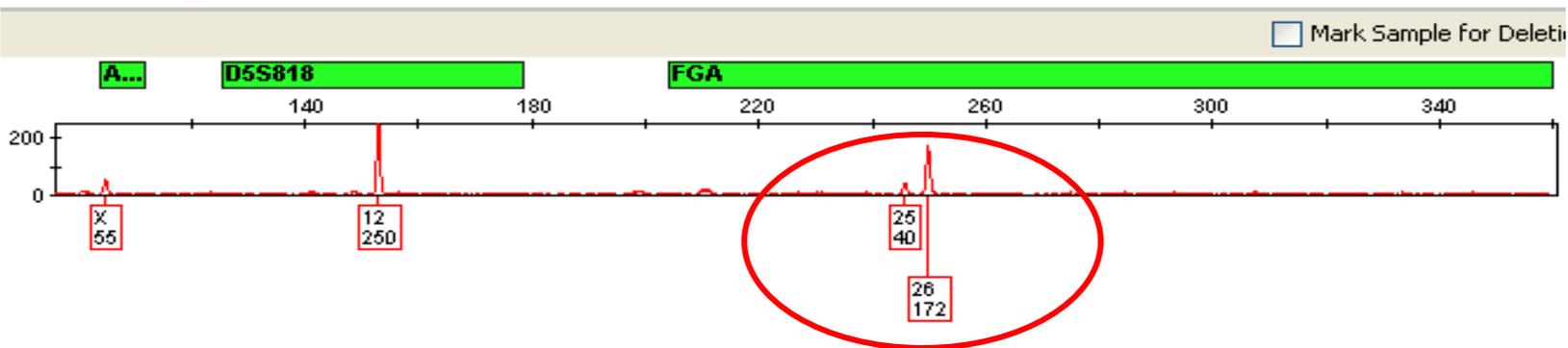
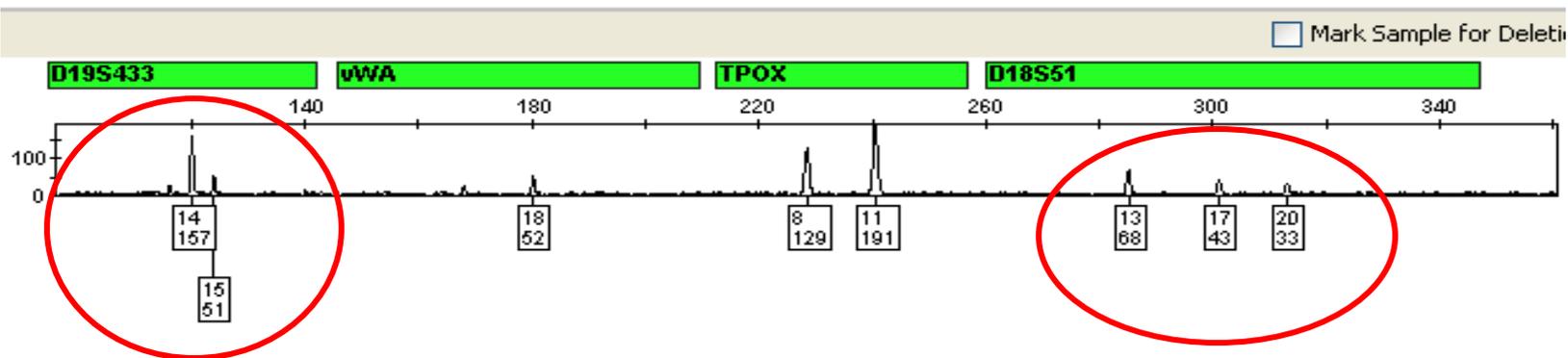
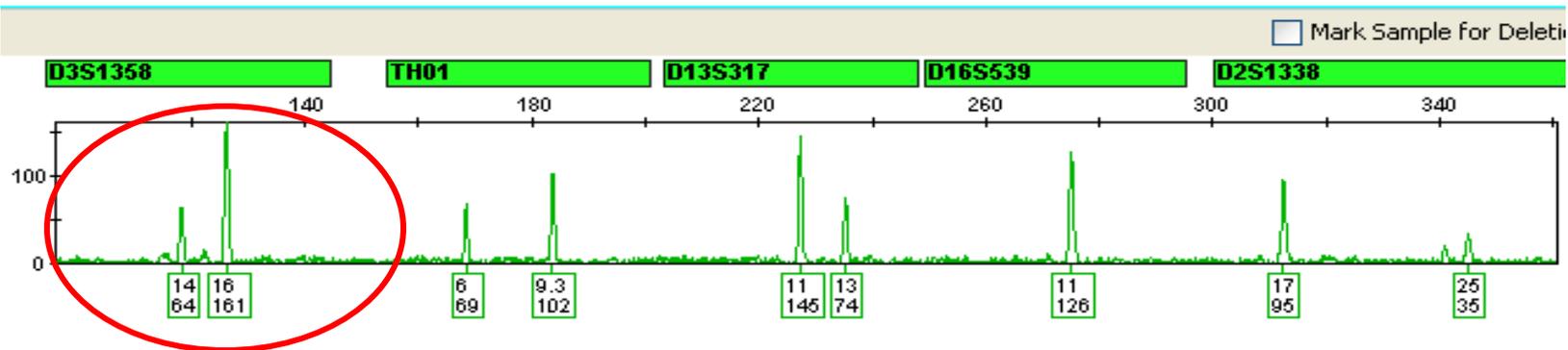
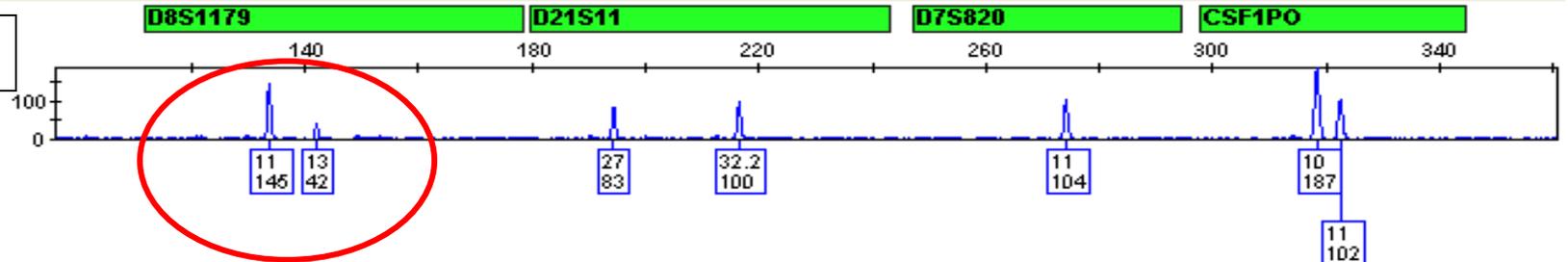
When to Consider Different Assumptions



- May need to consider multiple assumptions for data interpretation when:
 - Possible LT DNA profile
 - Stochastic effects (allelic drop-in, allelic drop-out, elevated stutter)
 - Possible minor contributor in mixed DNA profile
 - Possible known contributor(s) and deducing
 - More than 2 contributors (later today)

Profile 4

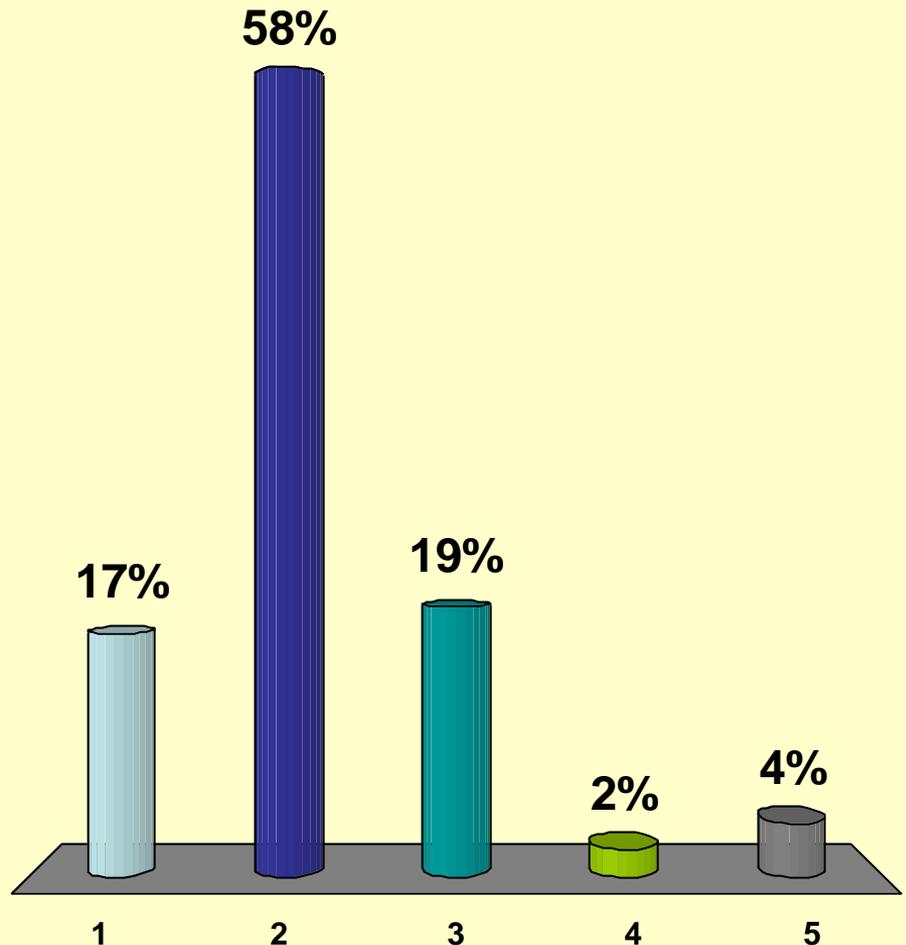
Single Source vs. Mixture?



I would report the previous profile as:

Data from 104 responses
ISHI Mixture Workshop (Oct 2012)

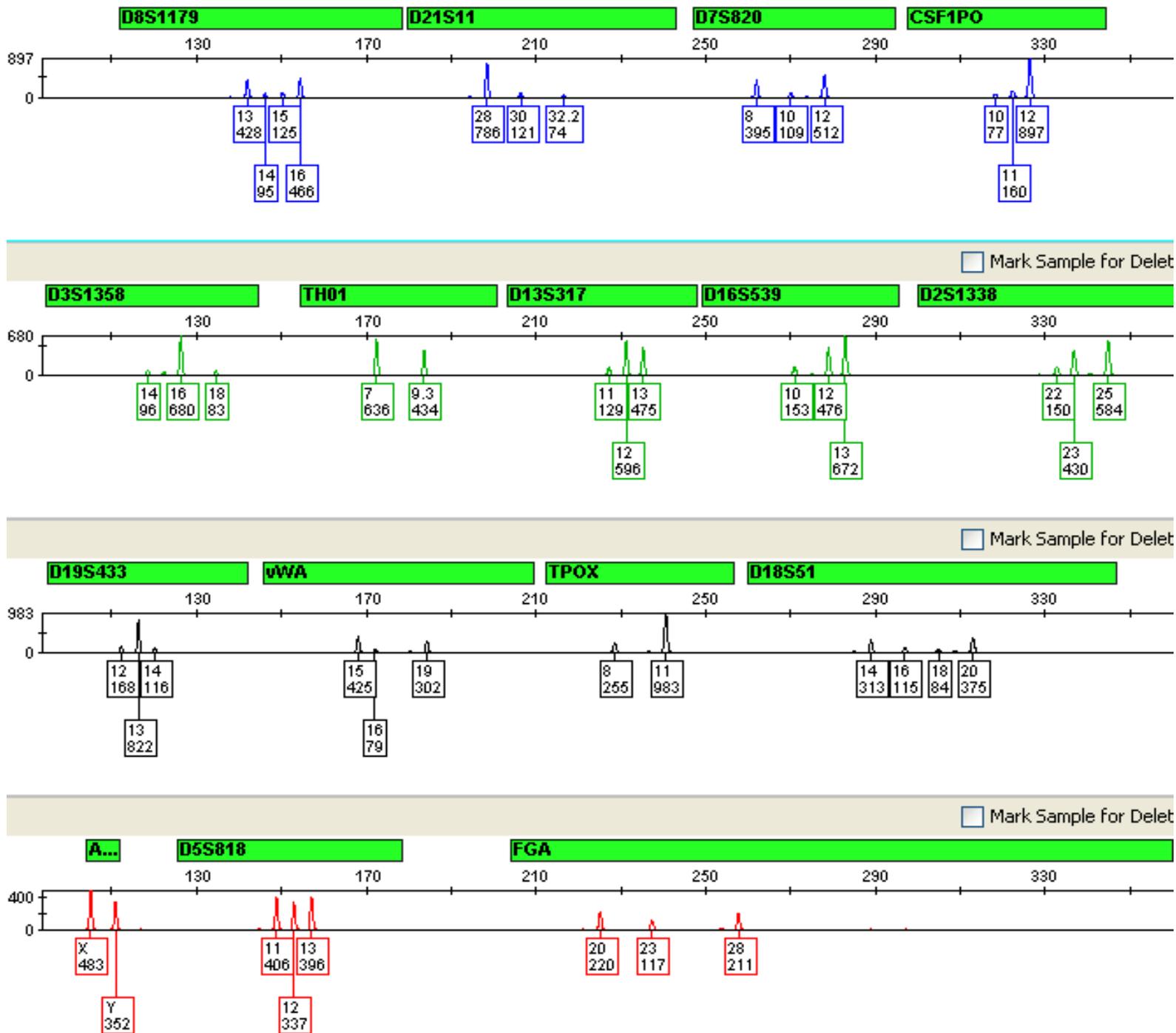
1. Single source
2. Mixture
3. Inconclusive
4. Single source and mixture
5. Not sure



Correct answer is a single-source sample from ~62 pg DNA template with drop-in at D18 (allele 20)

Profile 5

All Alleles vs. Allelic Drop-out? Two or More Contributors?



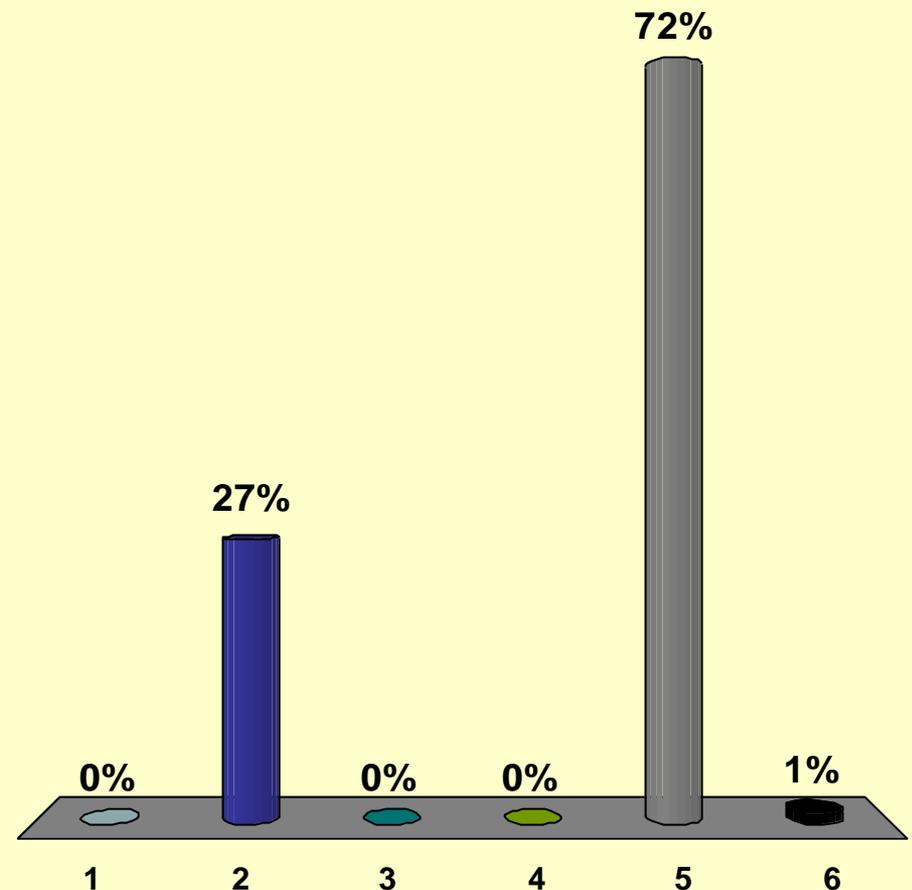
I am confident that the number of contributors in the previous profile is...

Data from 108 responses

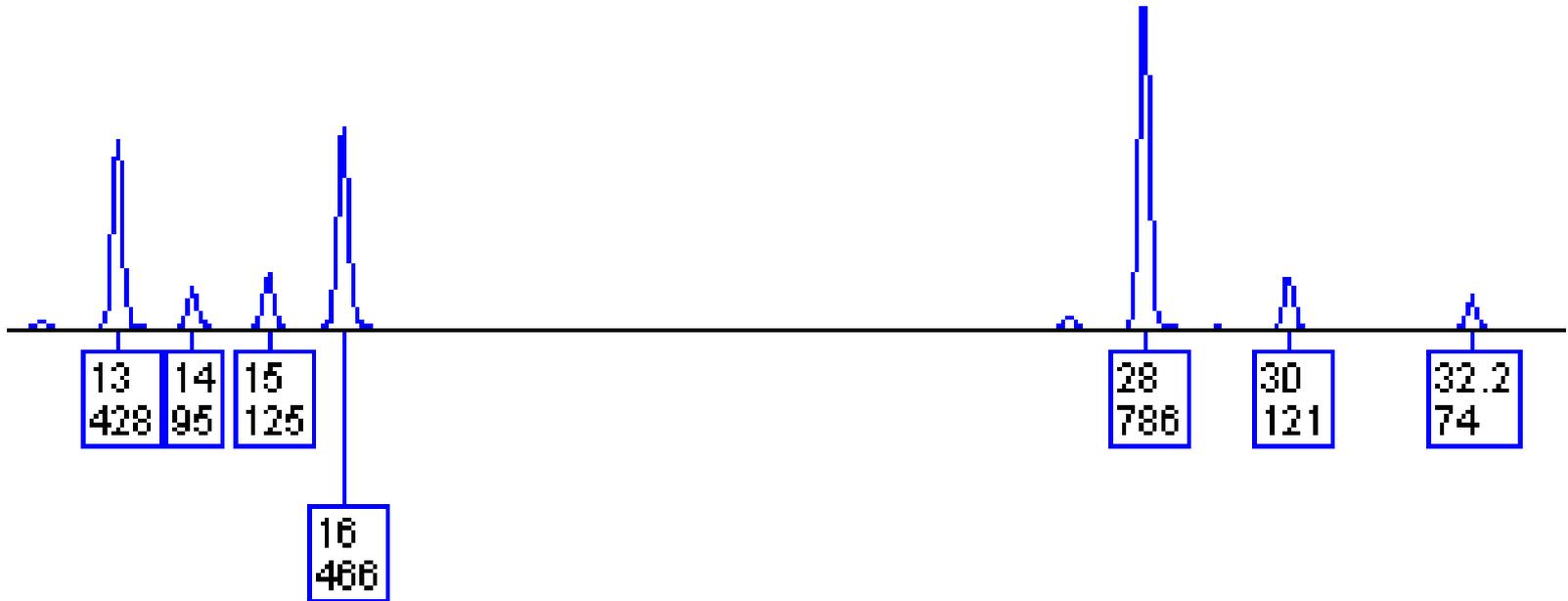
ISHI Mixture Workshop (Oct 2012)

1. 1
2. 2
3. 3
4. >3
5. At least 2
6. Not sure

*Correct answer is 2 contributors;
but "at least 2" is very appropriate*



Is Known Individual Included or Excluded?



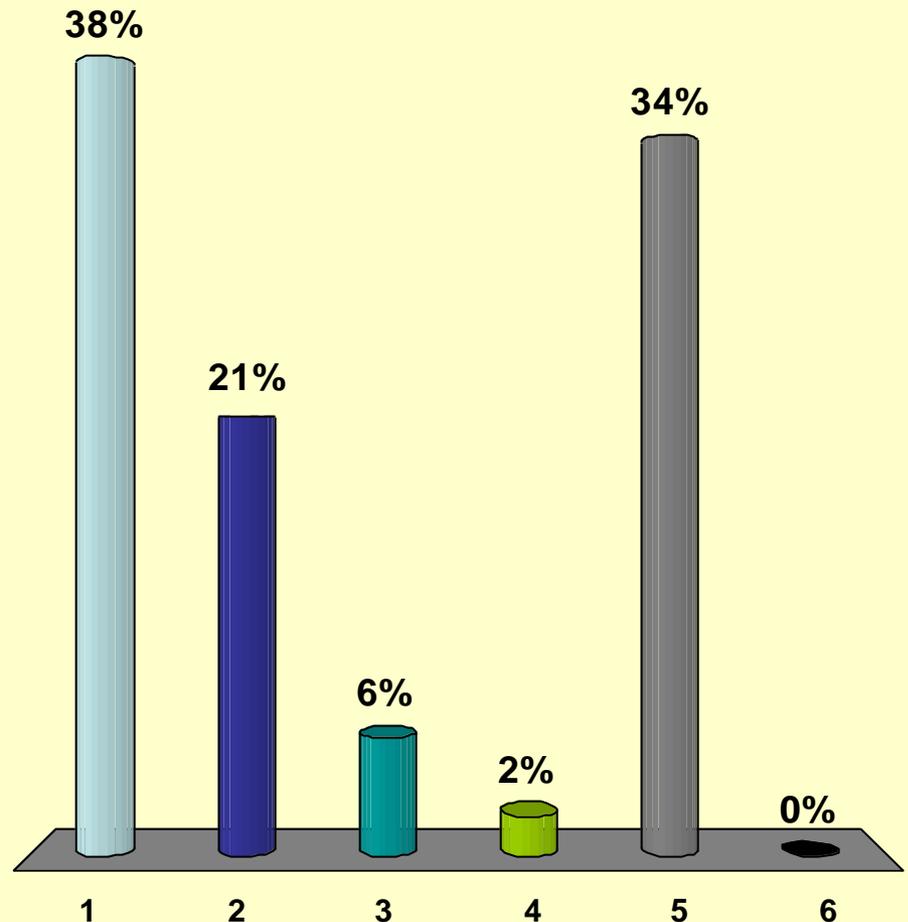
Known:
13,14

Known:
28,30

The known individual is:

Data from 106 responses
ISHI Mixture Workshop (Oct 2012)

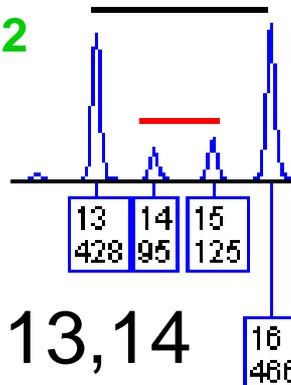
1. Included
2. Excluded
3. Inconclusive
4. Not sure
5. Included and excluded using different assumptions
6. Whatever my technical reviewer says!



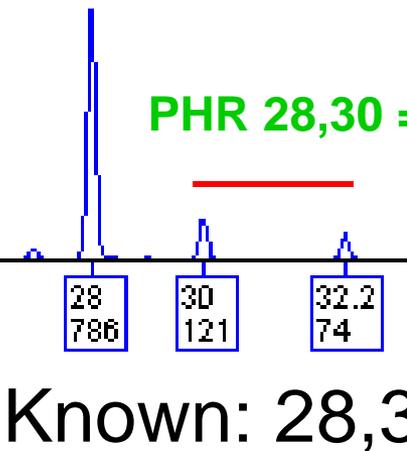
Is Known Individual Included or Excluded?



PHR 13,14 = 0.22



PHR 28,30 = 0.15 or 0.31



Known: 13,14

16
488

Known: 28,30

EXCLUDED as major & minor if assume only 2 contributors:

Must have 1 major and 1 minor contributor due to peak height ratio and mixture ratio calculations.

Major *must* have genotype of 13,16 and 28,28 and minor *must* have genotype of 14,15 and 30,32.2.

Genotypes NOT included even if alleles are!

Is Known Individual Included or Excluded?



Known: 13,14 16
466

Known:
28,30

NOT EXCLUDED based on alleles being present if assume >2 contributors

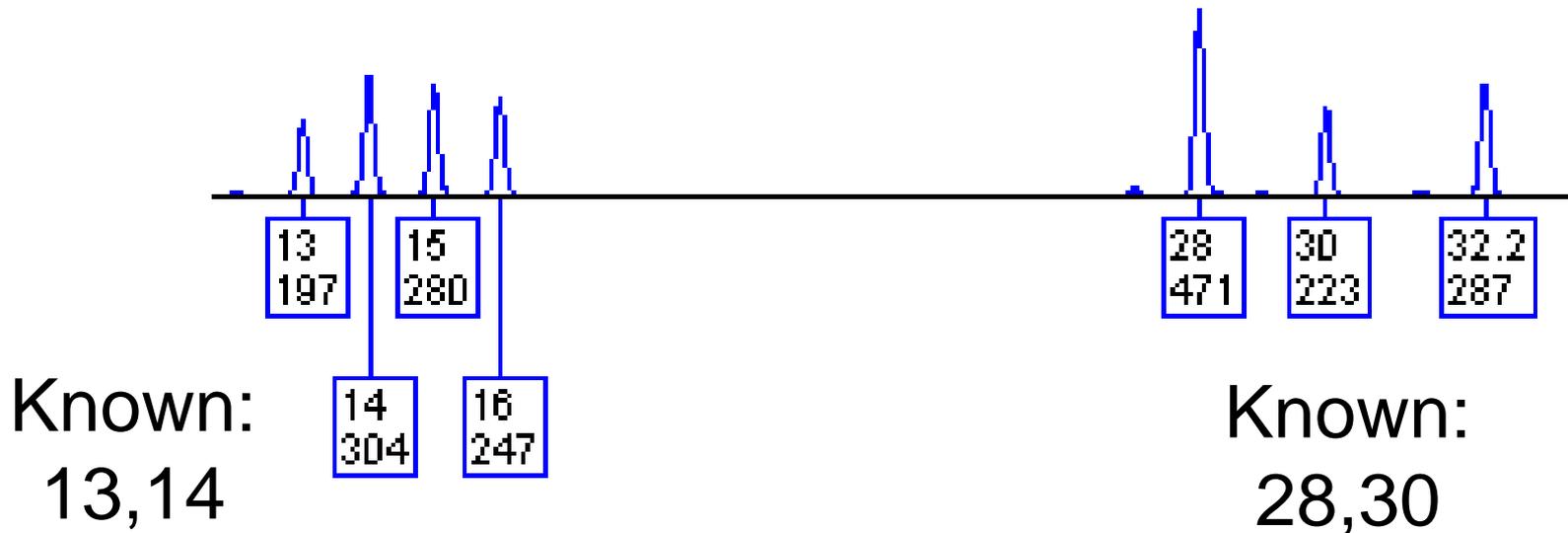
Assuming >2 contributors (≥ 2 minor) is reasonable due to the possibility of alleles missing since low peak heights (i.e., LT DNA) for alleles are observed at each locus

All alleles and genotypes represented?

Report: Inclusion (Stats??)

INCONCLUSIVE (insufficient data for minors)

Indistinguishable Mixture Profile



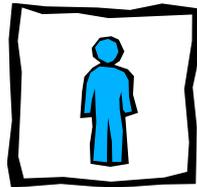
What if the genotypes **CANNOT** be distinguished?

Alleles are included, **BUT** are genotypes?

Reporting **Multiple Conclusions**

Different conclusions may result from using different assumptions.

If 2 contributors:



EXCLUDED

BUT

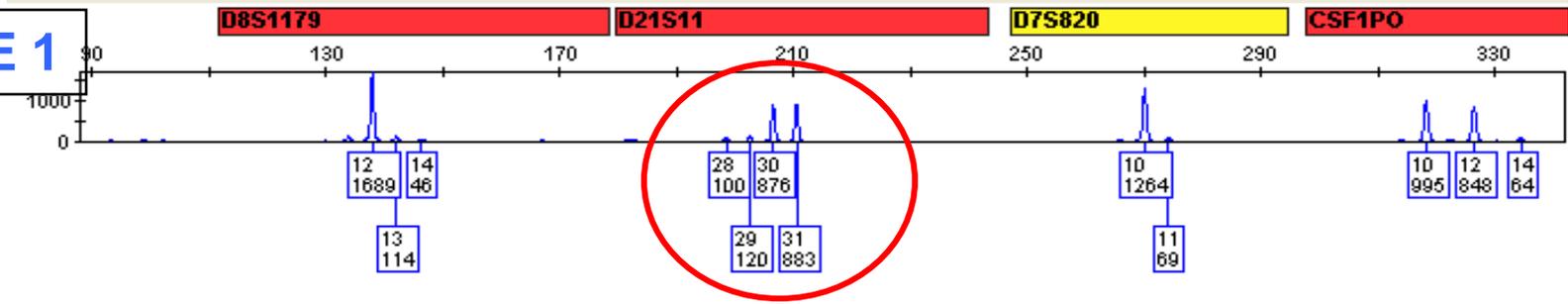
If 3 contributors:



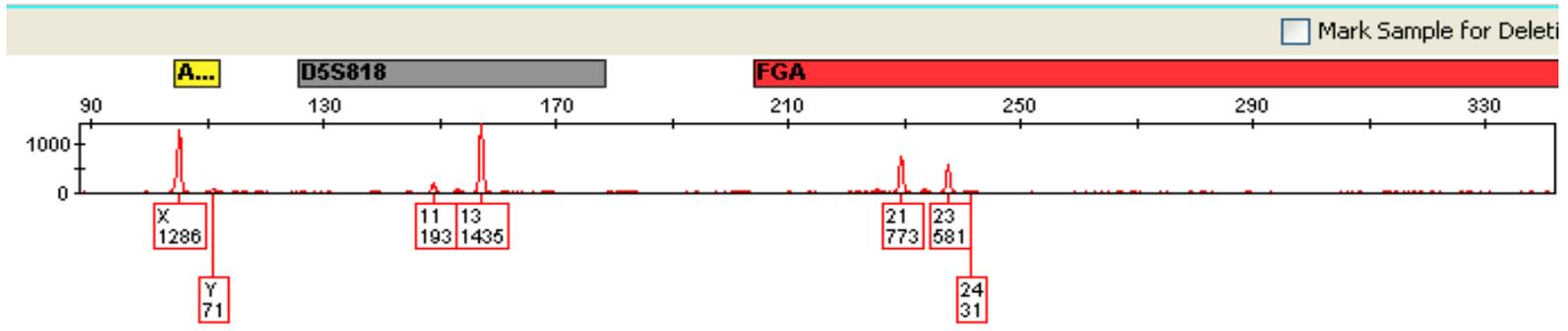
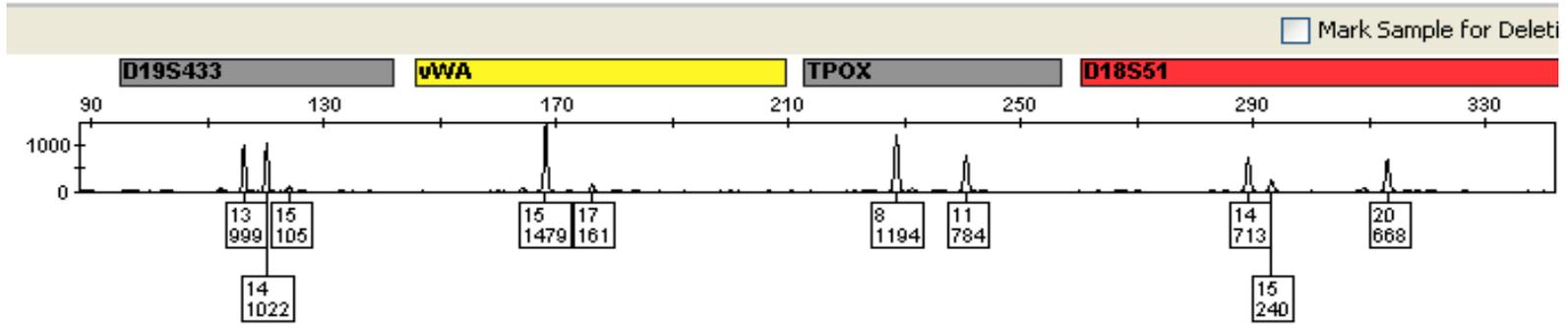
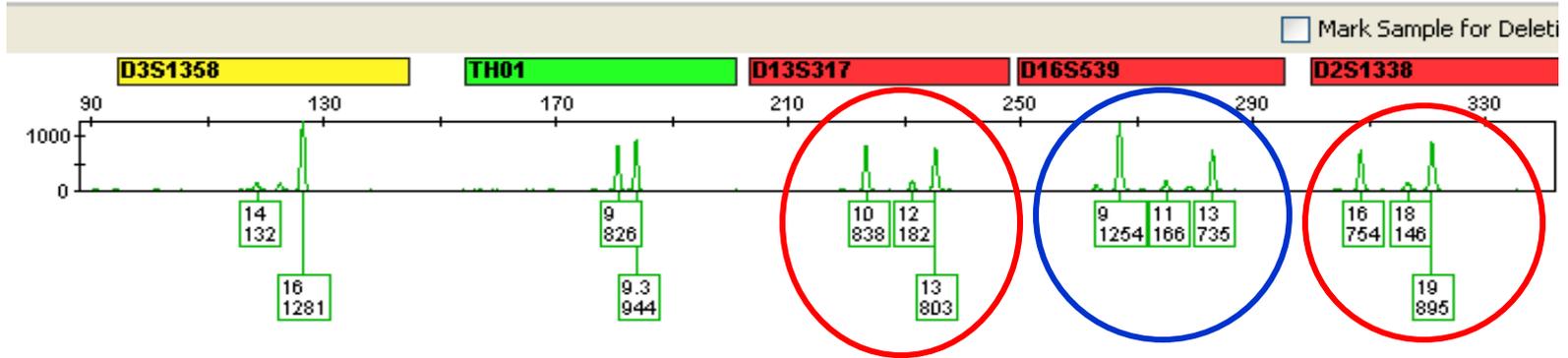
INCLUDED
INCONCLUSIVE

REPORT ALL CONCLUSIONS!

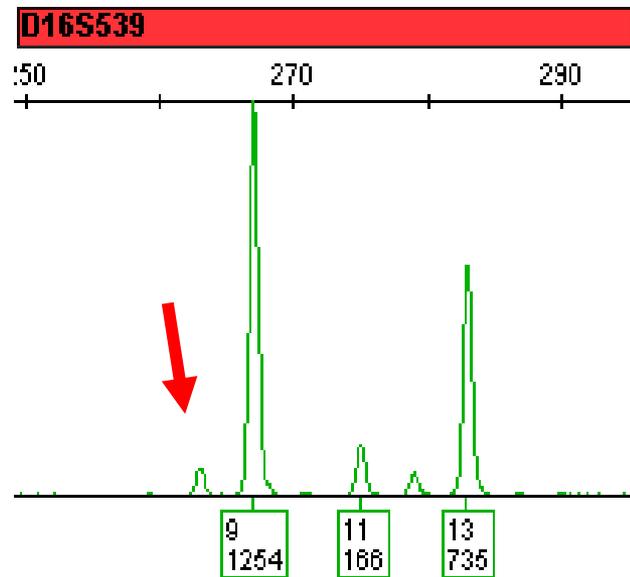
PROFILE 1



Stutter or true allele?
All alleles present?



PROFILE 1



8,11 = true minor contributor

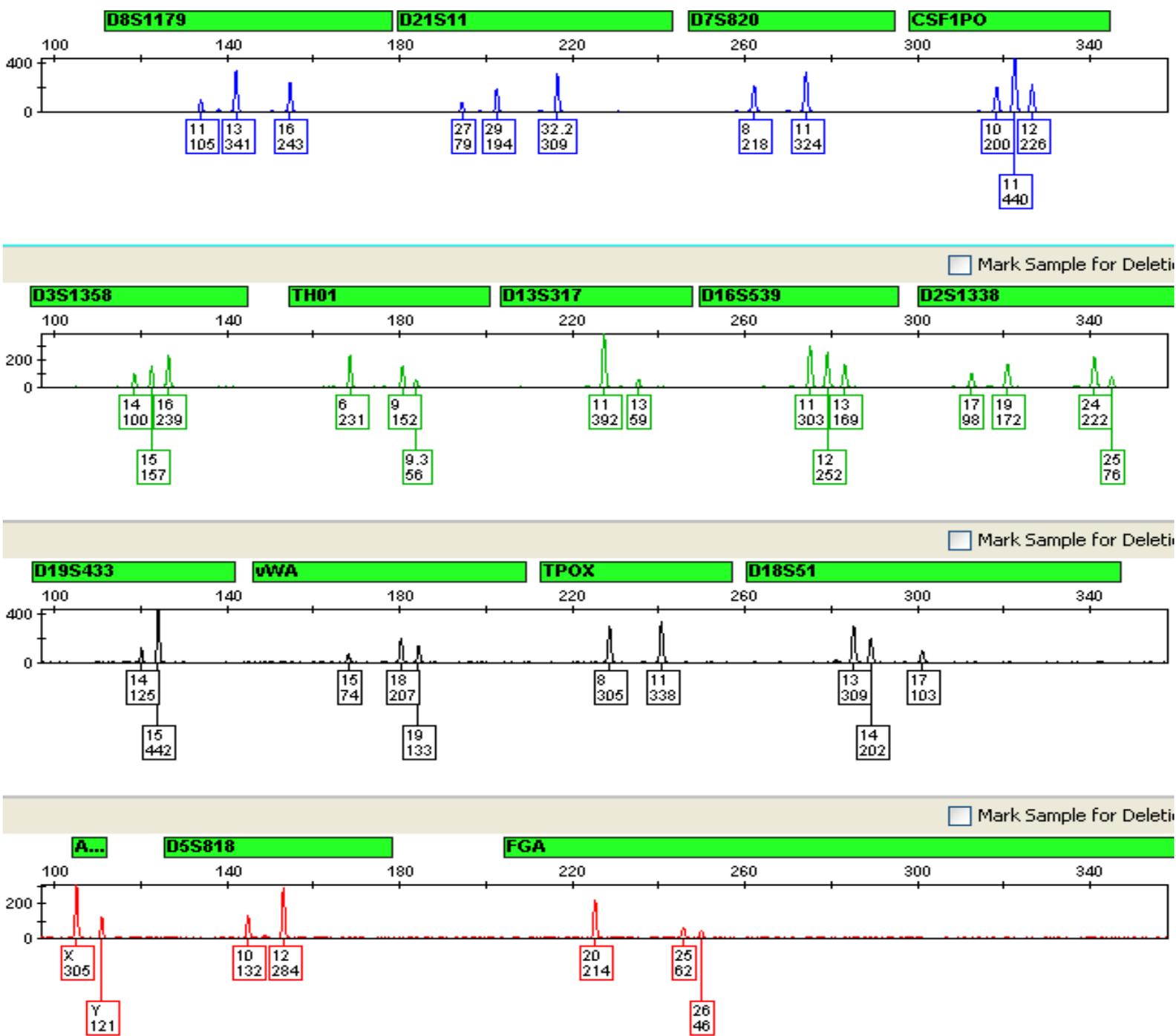
8 allele filtered out by software

Stutter or true allele?
All alleles present?

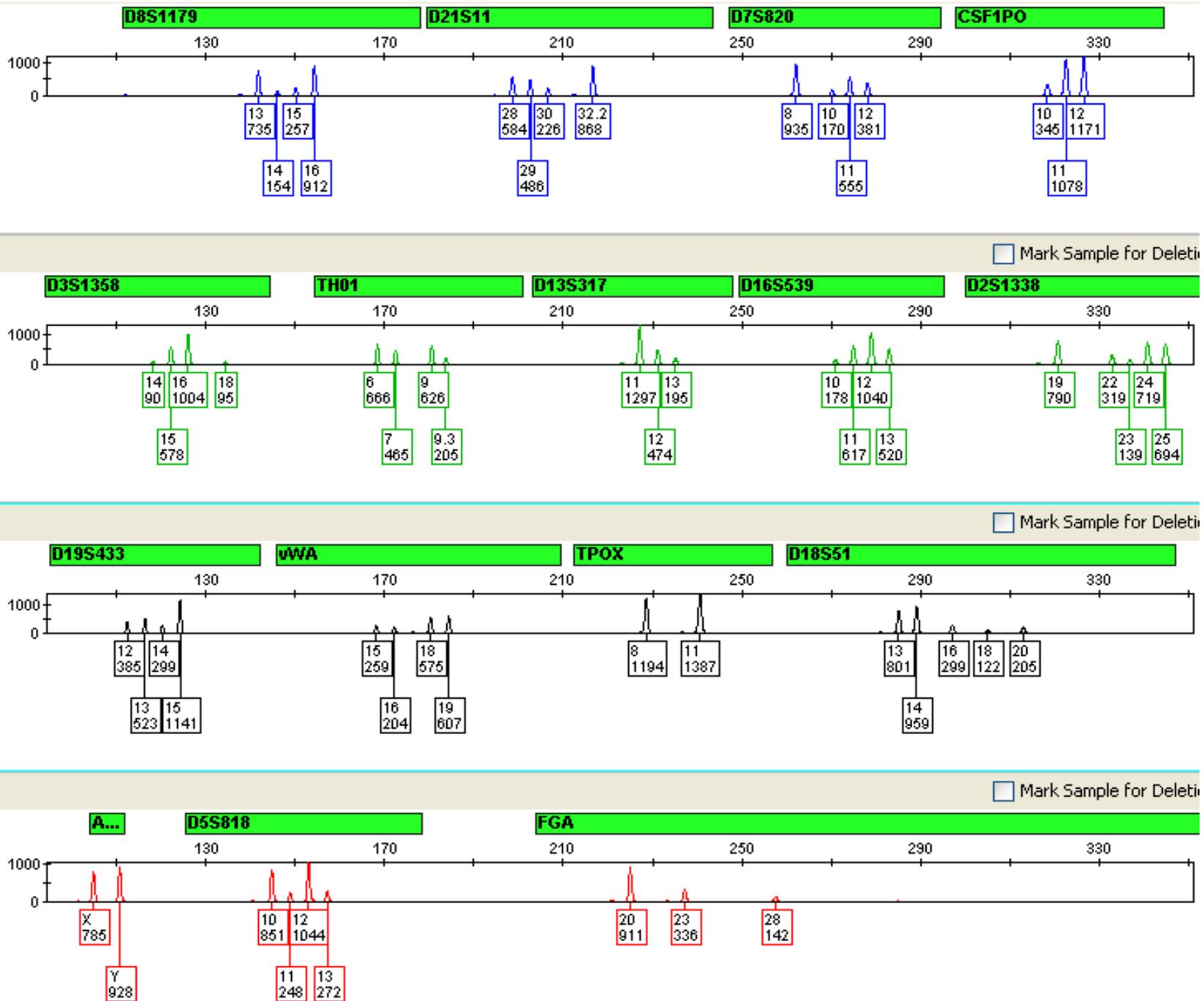
If assume 8 is a stutter peak and assume all peaks are present, would exclude the true contributor!

Uncertainty in evaluating the presence or absence of alleles leads to false inclusions and exclusions

Major vs. Indistinguishable? 2 contributors?



Major vs. Indistinguishable? Number of Contributors?



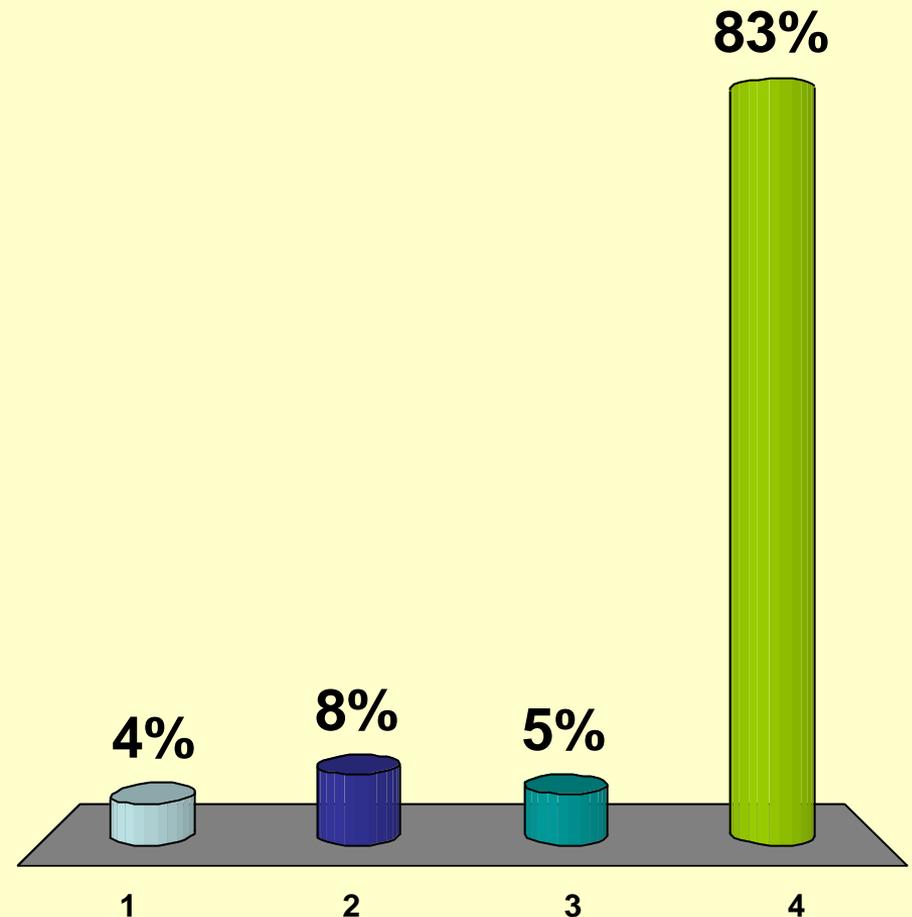
3 vs. 4 vs. 5 or more Contributors

Stay tuned for talk this afternoon

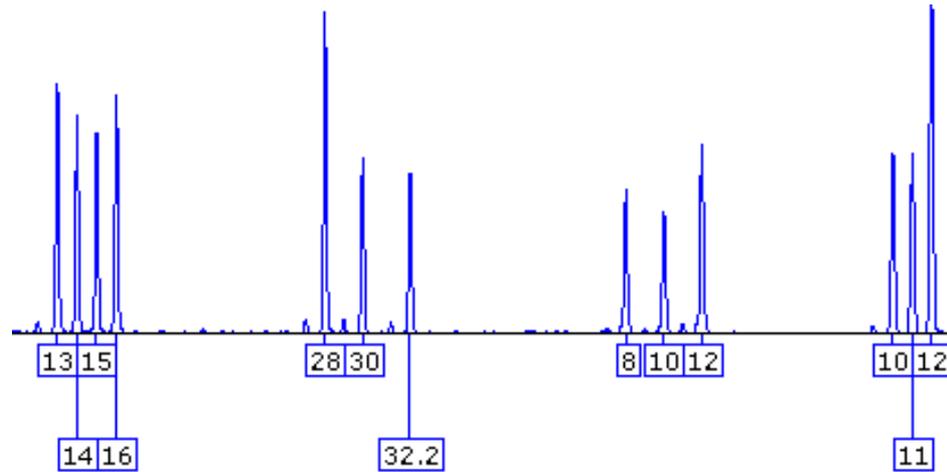
If a sample has more than one possible conclusion using different assumptions, then...

1. The best one for the client should be reported.
2. Some should be saved for court testimony.
3. Some should be ignored.
4. All should be reported.

Data from 92 responses
ISHI Mixture Workshop (Oct 2012)



Reporting Guidelines: **Two Inclusions**



Person 1



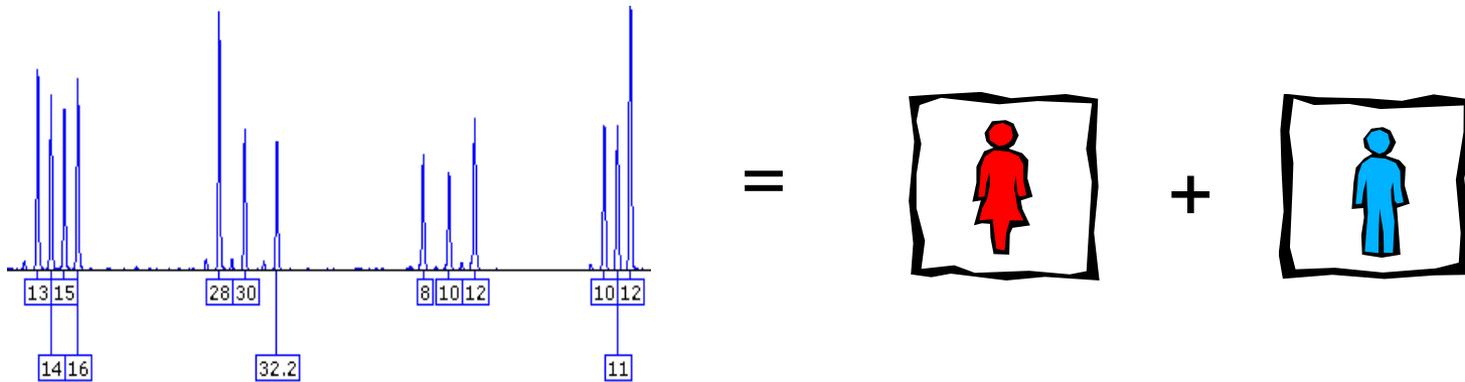
Included

Person 2



Included

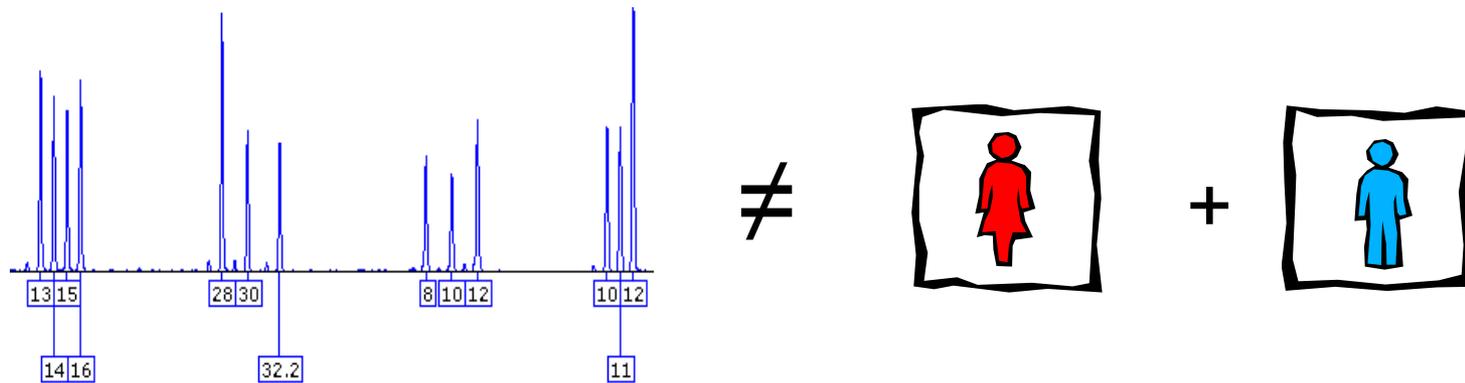
Two Inclusions



The results (all alleles and genotypes) **are consistent with a mixture** of DNA from the two individuals.

OR

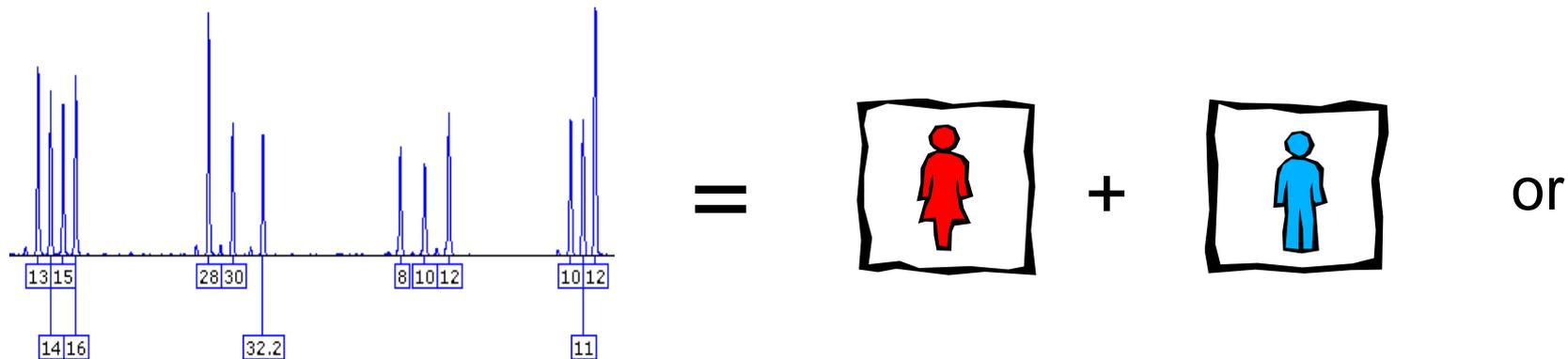
Two Inclusions



The **results are NOT consistent** with both of the individuals being contributors **together** in the sample.

This is important to report because....

Two Inclusions – Total of Four Possibilities



Could be:

- 2)  + ? or
- 3)  + ? or
- 4) ? + ?

Reporting



- Report all opinions, assumptions and conclusions
 - Single source or mixture
 - Number of contributors if mixture
 - Gender of contributor(s)
 - Partial profile (inconclusive loci)
 - Inclusions, Exclusions and Inconclusives
 - Statistics for **all** Inclusions
 - Explanations for Inconclusives
- Report results and conclusions for all data obtained for all samples

Reporting



- Consider the data from several scientific perspectives – for conclusions *and* statistical calculations
- Report all appropriate scientific conclusions and opinions in the laboratory report
- **ESPECIALLY** if the conclusions differ under different reasonable assumptions

Why Report?



- Opinions may be important to different individuals reading the report (e.g., law enforcement, prosecutor, defense attorney, client, judge, jury)
- Reports should be **neutral** to the case yet address the question(s) asked by the client

Why Report?



- Not all cases (<10%) make it to court
- Critical decisions often based on report and (mis)understandings alone
- If not provided in advance to all parties, opinions may not be admissible in court

THANK YOU!!